

Behavioral Responses of the Potato Psyllid (Hemiptera: Triozidae) to Volatiles from Dimethyl Disulfide and Plant Essential Oils

John Diaz-Montano · John T. Trumble

Revised: 25 May 2012 / Accepted: 18 July 2012
© Springer Science+Business Media, LLC 2012

Abstract The behavioral responses of the potato psyllid, *Bactericera cockerelli* (Sulc) to dimethyl disulfide (DMDS) and to 12 plant essential oils were examined in a glass Y-tube olfactometer. DMDS at doses of 0.5, 1, 2 and 5 g showed a significant repellent effect on the psyllids. Cedar wood, lime, savory, thyme and tea tree oils significantly repelled adults over a wide range of doses tested (1, 10, 100, 500, 1,000 and 2,000 μ l). The oils of clove and peppermint also had a significant repellent effect on adults at all doses except at the lowest dose (1 μ l). The residual effect of 1 g of DMDS persisted for 10 consecutive days whereas five oils (thyme, tea tree, peppermint, savory and clove) remained repellent for the 20-day longevity residual trial.

Keywords *Bactericera Cockerelli* · behavior · olfactometer · repellents

The potato psyllid, *Bactericera cockerelli* (Sulc) (Hemiptera: Triozidae), is a phloem-feeding pest of increasing concern in production of potato, *Solanum tuberosum* L., because its direct feeding causes retarded growth, chlorosis, and small tuber production (Al-Jabr and Whitney 2007). *Bactericera cockerelli* also causes economic losses to several other solanaceous vegetables including tomatoes, *Solanum lycopersicum* L. and peppers, *Capsicum* spp. (Liu and Trumble 2006; Munyaneza et al. 2007a, b; Teulon et al. 2009). In addition *B. cockerelli* has recently been found to transmit the bacterial pathogen “*Candidatus Liberibacter psyllaurous*” (a.k.a. *Ca. L. solanacearum*) that causes a newly emerging potato disease named Zebra Chip (ZC)

J. Diaz-Montano · J. T. Trumble
Department of Entomology, University of California, Riverside, CA 92521, USA

Present Address:

J. Diaz-Montano (✉)

Stored Product Insect Research Unit, USDA, Agricultural Research Service,
Center for Grain and Animal Health Research, 1515 College Av, Manhattan, KS 66502, USA
e-mail: john.diaz-montano@ars.usda.gov

(Hansen et al. 2008; Liefing et al. 2008; Liefing et al. 2009). Symptoms of ZC include internal brown discoloration in potato tubers and dark stripes and streaks when the tubers are fried (Munyaneza et al. 2007a, b, 2008, 2009; Munyaneza 2010). This disease has caused millions of dollars in losses to the potato industry in the United States (Munyaneza et al. 2007a, b, 2009) as a result of potato yield losses, which can exceed 80 % (Munyaneza et al. 2008). Currently, the control of this bacterial disease depends on the management of *B. cockerelli*, which relies on the use of chemical insecticides (Liu and Trumble 2005; Goolsby et al. 2007; Gharalari et al. 2009). Chemical control is expensive, can reduce populations of natural enemies, and has led to the development of resistance (Liu and Trumble 2007). One alternative is to investigate plant essential oils that have proven useful in other plant-pest systems (Adorjan and Buchbauer 2010; Nerio et al. 2010). Therefore, we chose to explore the use of plant essential oils and other compounds as repellents of *B. cockerelli*.

Numerous plant essential oils from different species have been tested to assess their repellent and insecticidal effects to various pests such as mosquitoes, cockroaches, flies, lice and mites (Adorjan and Buchbauer 2010; Nerio et al. 2010). Plant essential oils have also been examined on insect pests of agricultural importance. Essential oil from *Thymus persicus* L. was examined against two stored-product beetles *Tribolium castaneum* (Herbst) and *Callosobruchus maculatus* (Fabricius) and a repellency of 70.4 % and 82.4 %, respectively was observed (Moharrampour et al. 2008). The repellent effect of essential oils of thyme, *Thymus vulgaris* L., patchouli, *Pogostemon cablin* (Blanco) Benth., and lemon-scent gum, *Corymbia citriodora* (Hook.) K. D. Hill and L. A. S. Johnson, on *Bemisia tabaci* (Gennadius) biotype B was studied in choice test experiments and all three had a significant effect on *B. tabaci* (Yang et al. 2010a). Yang et al. (2010b) tested the repellency of four biorational insecticides including Bugoil, a mixture of canola oil, thyme oil, tagetes oil (*Tagetes erecta* L.) and wintergreen oil (*Gaultheria procumbens* L.), and found that Bugoil provided a substantial repellency to *B. cockerelli* adults and deterred oviposition. Nissinen et al. (2005) studied the effect of limonene (a monoterpene in carrot (*Daucus carota* L.) essential oil) on carrot psyllids, *Trioza apicalis* Förster, and this material significantly increased the number of eggs laid by psyllids. However, in other studies, limonene was shown to have a repellent effect on carrot psyllids (Nehlin et al. 1994; Valterová et al. 1997; Kainulainen et al. 2002). Mann et al. (2011) studied the effect of garlic chive essential oil (*Allium tuberosum* Rottler) compared with clean air and citrus leaves in a T-olfactometer and it repelled Asian citrus psyllid, *Diaphorina citri* Kuwayama, adults. Koschier and Sedy (2003) observed a significant repellent effect of the essential oil of rosemary (*Rosmarinus officinalis* L.) to onion thrips, *Thrips tabaci* Lindeman, adult females in a glass Y-tube olfactometer.

Although relatively few essential oils have proven successful against horticultural pests, and only a few of them are used in agriculture (Isman 2006), these natural products have the potential to provide efficient and safe repellents for humans, the environment (Nerio et al. 2010) and beneficial insects (Egigu et al. 2010). There are very few studies related to the repellent effect of plant essential oils and other compounds to psyllids of agricultural importance. Our objectives were to test if the olfactometer system was an effective tool in eliciting and quantifying a behavioral response of *B. cockerelli* adults to odors and subsequently to evaluate the repellent effect of volatiles from dimethyl disulfide and different plant essential oils to *B. cockerelli* adults.

Materials and Methods

Insect Culture and Plant Material *Bactericera cockerelli* populations used in these experiments were originally collected from fresh market tomatoes in Weslaco City (Hidalgo County, Texas, USA) in June 2005 and were maintained on tomato ‘Yellow Pear’ plants under greenhouse conditions at 20–30 °C and 20–40 % RH, with a photoperiod of 14:10 (L:D) h at the University of California, Riverside, Department of Agricultural Operations. Tomato plants were grown in pots (10.0 cm in diameter by 10.0 cm in height) with one plant per pot, and replaced as necessary. Insects used in the experiment were taken from the colony ~1 h before the initiation of the tests.

Olfactometer System The behavioral responses of *B. cockerelli* adults to volatiles from tomato, potato, and pepper leaves and from dimethyl disulfide and various essential oils, were studied using a glass Y-tube olfactometer. The olfactometer had a 1.3 cm inner diameter, 12 cm length from the base of the tube to the Y-junction, and two 13 cm length arms at an angle of 45° with the ends reduced to 0.6 cm inner diameter. The experimental approach was partially based on the methods described by Koschier et al. (2000) and Davidson et al. (2008). Experiments were carried out in a room at 20–24 °C and 40–45 % RH. Four fluorescent tubes fixed above the device provided illumination. The Y-tube was positioned vertically (90°) and was connected at the base to a suction pump. Distal ends of the two arms were individually connected to a filtering (Erlenmeyer) flask (Pyrex® 5340-500, Corning Inc., Corning, NY; 500 ml glass narrow mouth 3.5 cm in diameter by ~18 cm in height, with a side arm tubulation 10 mm outer diameter) by means of a rubber stopper (3 and 3.5 cm in diameter, lower and upper part respectively by 2.4 cm in height) with a glass tube (1 cm in diameter by ~10 cm in height) inserted in the center of the stopper. The air passed through a charcoal filter (Activated Charcoal Trap, 36 cm in length by 2.7 cm in diameter glass tube) connected to the silicone tubing and the two arms of the Y-tube. Airflow was regulated to 1.97 mm/s (0.12 liter/min) using two airflow meters (Cole-Parmer®, Vernon Hills, IL), one for each arm. All connections between the pump, the airflow meter, the Y-tube, the two Erlenmeyer flasks and the charcoal filter were made with rigid silicone tubing (6 mm inner diameter). The air was drawn through the Y-tube for at least 1 min before introducing the *B. cockerelli* adults.

Leaf Volatiles These experiments were performed in order to establish whether or not the Y-tube olfactometer system was an effective tool in eliciting and quantifying a behavioral response of *B. cockerelli* adults to odors. The varieties of pepper, potato and tomato used were ‘California Wonder’, ‘Calwhite’ and ‘Yellow Pear’, respectively. Plants used in these experiments were 3–4 weeks old and grown in greenhouse conditions at 20–30 °C and 20–40 % RH, with a photoperiod of 14:10 (L:D) h.

Initially, experiments with 5, 10 and 15 g of pepper, potato and tomato leaves were conducted. Whole leaves (pepper) and leaflets (tomato and potato) were placed inside one of the Erlenmeyer flask; the other flask was left empty as a ‘blank’. This test was designed to determine if attractiveness varied with the amount of leaf material, and if so, how much leaf material should be used.

We performed the trials using the basic experimental design used previously (Borges et al. 1998; Hardie et al. 1990; Gökçe et al. 2005; Rodriguez-Saona et al.

2006; Turlings et al. 2004), where insects were released in groups rather than singly. In our preliminary trials, group releases provided better statistical separation with fewer experiments and less time required than releases of individual insects (data not shown). Subsequently psyllid adults were released at the rate of 50 at a time, with 100 psyllid adults released per each experiment. The Y-tube olfactometer was alternated 180° between replicates. This experimental design (2 replicates of 50 psyllid adults) was used for all subsequent tests. One concern was that the males were simply attracted to females, and the preference data would be enhanced if the males followed the females rather than selecting an arm on the basis of the odors presented. While there have been pheromones identified for other psyllid species, there have been no pheromones documented for *B. cockerelli* (Guedot et al. 2009). Therefore, a separate experiment was performed in order to test if there were any differences between male and female attraction to leaf volatiles (using 15 g tomato leaves) with only one gender present. Leaves were placed inside one of the Erlenmeyer flask; the other flask was left empty as a 'blank'. All other conditions were as previously reported.

Dimethyl Disulfide (DMDS) Dimethyl disulfide has been shown to be repellent to a related psyllid (*D. citri*) (Onagbola et al. 2011; Mann et al. 2011) and was considered a candidate for potato psyllid control. Dimethyl disulfide is found commercially incorporated into SPLAT (Specialized Pheromone and Lure Application Technology), which is a slow release wax formulation (DMDS; 'SPLAT', ISCA Technologies, Riverside, CA). In order to test the response of *B. cockerelli* adults to SPLAT-DMDS, doses of 0.5, 1, 2 and 5 g were compared first against a blank arm. Then, SPLAT-DMDS (1 g) was compared against to an arm with 15 g of tomato or potato leaves. Finally, SPLAT-DMDS (1 g) + tomato leaves (15 g) in one arm was compared against tomato leaves (15 g) in the other arm.

In addition, the residual effect of 1 g of SPLAT-DMDS to *B. cockerelli* adults against a blank arm was tested for 20 consecutive days. In between trials (20 day-period) the material was held in the Erlenmeyer flask containing the SPLAT-DMDS and was left open to ventilate in a laboratory fume hood. A total of 100 psyllid adults were used for each experiment as described in the previous experiment.

Essential Oils A total of 12 essential oils (Table 1) were used to study the repellent effect of six doses (1, 10, 100, 500, 1,000 and 2,000 µl) to *B. cockerelli* adults. The oil was placed on a filter paper (9 cm diameter) for all experiments. Additionally, the residual effect of 1,000 µl of the essential oils with the highest repellent effect was tested for 20 days. After each day the Erlenmeyer flask containing the oil was left open to ventilate in a laboratory fume hood.

Another experiment was performed to observe if there were any differences between male and female repellency to essential oils. In our preliminary experiments, clove (*Syzygium aromaticum* L) oil showed one of the strongest repellency effects to *B. cockerelli* adults; therefore it was chosen for this experiment. Clove oil (500 µl clove oil) was placed inside one of the Erlenmeyer flask; the other flask was left empty as a 'blank'. There were 2 replicates of 50 psyllid adults for each male and female experiment.

Essential Oils with Leaves These experiments were conducted in order to observe if the presence of tomato leaves from 'Yellow Pear' interferes with the repellent effect

Table 1 List of essential oils studied for their repellency to *B. cockerelli* adults

Essential Oil	Scientific name (Family)	Provider company
Cassia	<i>Cinnamomun cassia</i> Blume (Lauraceae)	SAFC Supply Solutions. St Louis, MO
Cedar Wood	<i>Juniperus virginiana</i> L. (Cupressaceae)	Tokyo Kasei Kogyo Co., Ltd. Tokyo, Japan
Celery	<i>Apium graveolens</i> L. (Apiaceae)	Herb Pharm. Williams, OR
Bud Clove	<i>Syzygium aromaticum</i> L. (Myrtaceae)	SAFC Supply Solutions. St Louis, MO
Garlic	<i>Allium sativum</i> L. (Liliaceae)	Sigma-Aldrich Co. St Louis, MO
Lime	<i>Citrus aurantifolia</i> (Christm.) Swingle (Rutaceae)	SAFC Supply Solutions. St Louis, MO
Patchouli	<i>Pogostemon cablin</i> Benth. (Labiatae)	SAFC Supply Solutions. St Louis, MO
Peppermint	<i>Mentha piperita</i> L. (Lamiaceae)	Sigma-Aldrich Co. St Louis, MO
Rosemary	<i>Rosmarinus officinalis</i> L. (Labiatae)	SAFC Supply Solutions. St Louis, MO
Savory	<i>Satureja hortensis</i> L. (Labiatae)	Spectrum® Quality Products. INC. Gardena, CA
Tea Tree	<i>Melaleuca alternifolia</i> (Maiden & Betche) Cheel (Myrtaceae)	Spectrum Chemical MFG Corp. Gardena, CA
Thyme	<i>Thymus vulgaris</i> L. (Labiatae)	SAFC Supply Solutions. St Louis, MO

of essential oils, which more closely relates to what would happen in a field or greenhouse situation. Tomato leaves (10 g) were combined, in separate experiments, with each oil at different doses (1, 10, 100 and 500 μ l) inside one of the Erlenmeyer flask; the other flask was left blank. Complementary experiments were conducted, except that the other flask contained tomato leaves (10 g). There were 2 replicates of 50 psyllid adults for all experiments.

Statistical Analyses For the olfactometer experiments, the responses of *B. cockerelli* adults to leaves, control (blank), DMDS, essential oils or essential oils with leaves for each arm of the olfactometer system were compared by the chi-square test for goodness-of-fit ($\alpha=0.05$) by using PROC FREQ (SAS Institute 2003).

Results

Leaf Volatiles When 50 *B. cockerelli* adults were released at the same time significantly more psyllids preferred the arm containing the pepper, potato and tomato leaves in all the experiments (Table 2). Therefore all subsequent trials used groups of adults. *Bactericera cockerelli* males or females tested separately were significantly more attracted to the arms with tomato leaves than to blank arms (Table 2). Therefore, we were confident that mixed *B. cockerelli* populations of male and female adults provided an accurate odor response.

Dimethyl Disulfide (DMDS) All the DMDS doses (0.5, 1, 2 and 5 g) tested against the blank arms had a significant repellent effect on *B. cockerelli* adults (Fig. 1a, b, c and d). The repellent effect varied from 62 to 81 %.

When 1 g of DMDS was in one arm of the Y-tube olfactometer system (without leaves) and challenged against potato leaves (15 g) or tomato leaves (15 g) in the

Table 2 Attractiveness of *B. cockerelli* adults (released in a Y-tube olfactometer system) to pepper, potato, and tomato leaf volatiles

Trt.	<i>B. cockerelli</i> Response ^a		χ^2	<i>P</i>
	Blank	Leaves		
Tomato (5 g)	39	61	4.84	0.0278 ^b
Tomato (10 g)	38	62	5.76	0.0164 ^b
Tomato (15 g)	33	67	11.56	0.0007 ^b
Pepper (15 g)	40	60	4.0	0.046 ^b
Potato (15 g)	29	71	17.64	0.00003 ^b
Tomato (15 g) ^c	36	64	7.84	0.0051 ^b
Tomato (15 g) ^d	32	68	12.96	0.0003 ^b

^a 100 *B. cockerelli* adults tested per experiment

^b Significantly more *B. cockerelli* adults preferred the arm with leaves ($P < 0.05$)

^c Only *B. cockerelli* male adults

^d Only *B. cockerelli* female adults

other arm, DMDS significantly repelled *B. cockerelli* adults (Fig. 2a and b). Furthermore, 1 g of DMDS along with tomato leaves (15 g) in one arm of the Y-tube olfactometer system compared against tomato leaves (15 g) in the other arm also significantly repelled *B. cockerelli* adults (Fig. 2c).

The residual effect of DMDS (1 g) on *B. cockerelli* adults was significant for 10 consecutive days. The percentage of repellency ranged between 45 and 65 % and the average was 59.8 %. From the 11th to the 15th day the effects varied and there were no significant effects in the last 5 days (Fig. 3a).

Essential Oils

Repellency of Plant Essential Oils Against Blank Arms Patchouli oil had no effect at any dose on *B. cockerelli* adults (Table 3). Cassia oil had a repellent effect only at two doses (500 and 2,000 μl). The oils of garlic, celery and rosemary had a repellent effect on potato psyllid adults at all doses except the two lowest doses (1 and 10 μl). Another 2 essential oils (clove and peppermint) had a repellent effect on adults at all doses except at the lowest dose (1 μl). The essential oils of cedar wood, thyme, tea tree, lime and savory had a repellent effect on *B. cockerelli* adults at all 6 doses (1, 10, 100, 500, 1,000 and 2,000 μl) (Table 3).

When 500 μl of clove oil were used to separately test the repellency of populations of *B. cockerelli* males and females, significantly more adults were repelled regardless of gender (Table 3). This further supported the use of mixed genders in the trials.

Residual Effect of Essential Oils The seven essential oils with the highest repellent effect on *B. cockerelli* adults observed in the previous experiment were tested daily for 20 days to observe their residual effect on *B. cockerelli*. The residual effect of cedar wood oil on *B. cockerelli* adults was significant for the first 2 days only and the

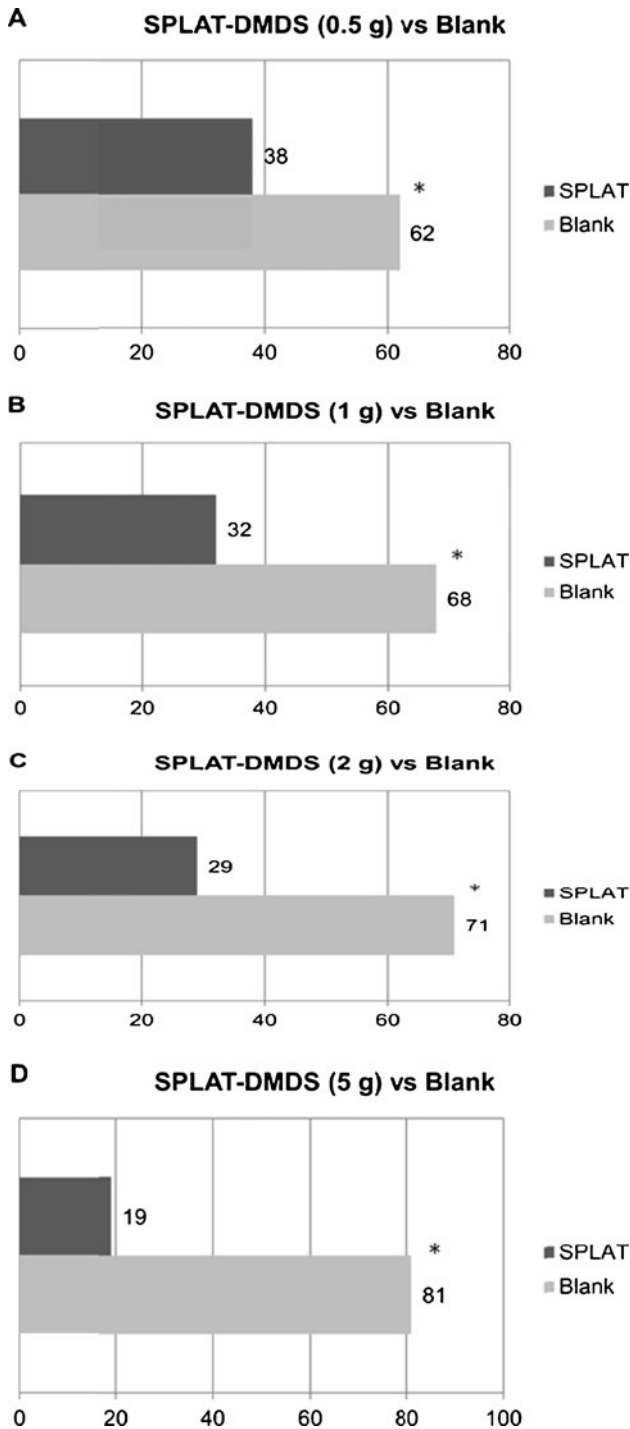


Fig. 1 Repellent effect of SPLAT-DMDS doses on *B. cockerelli* adults. **a.** SPLAT-DMDS (0.5 g) vs Blank. **b.** SPLAT-DMDS (1 g) vs Blank. **c.** SPLAT-DMDS (2 g) vs Blank. **d.** SPLAT-DMDS (5 g) vs Blank. An * indicates a significant difference at $P < 0.05$, chi-square test for goodness-of-fit

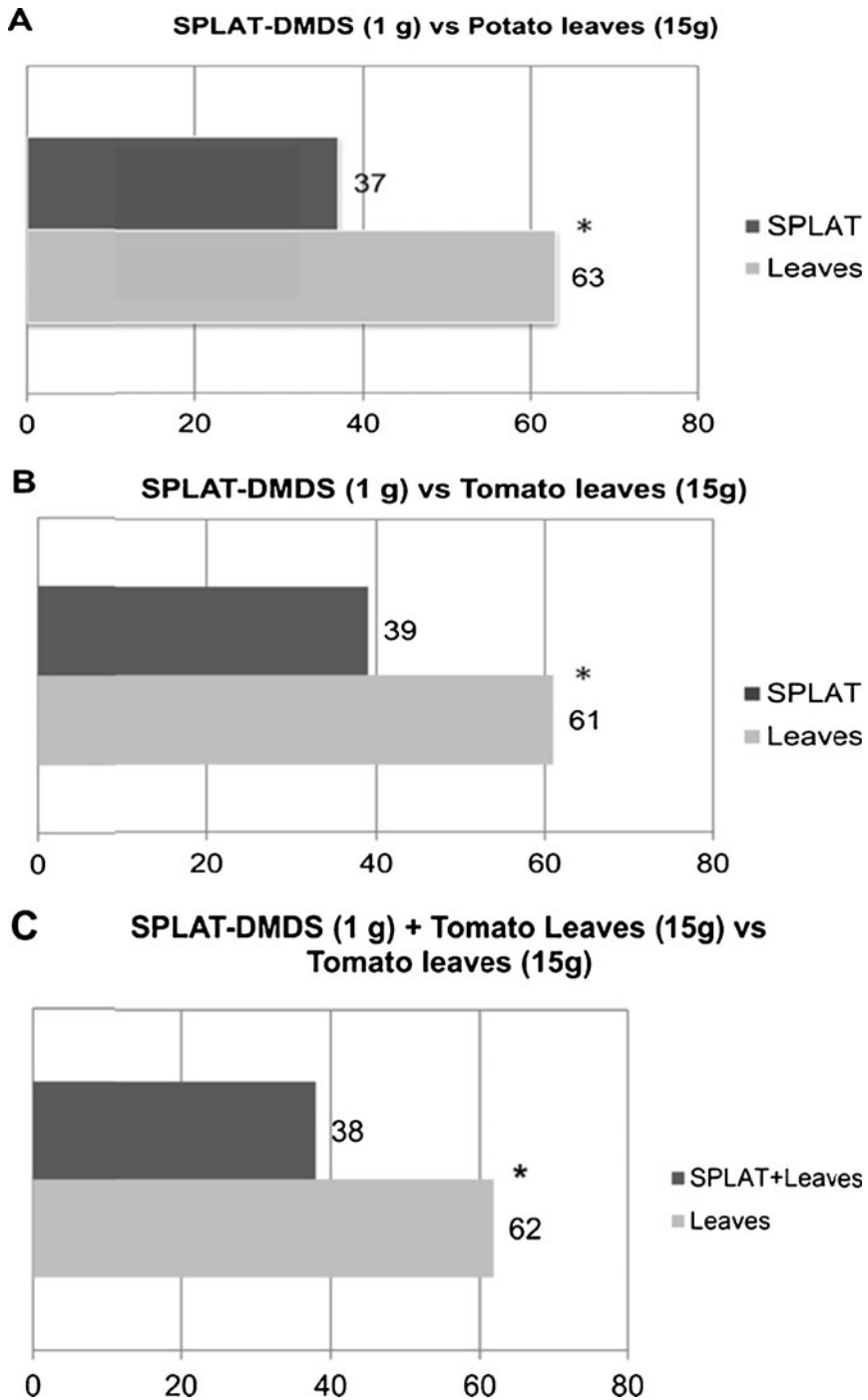


Fig. 2 Repellent effect of SPLAT-DMDS doses on *B. cockerelli* adults with the presence of leaf volatiles. **a.** SPLAT-DMDS (1 g) vs Potato leaves (15 g). **b.** SPLAT-DMDS (1 g) vs Tomato leaves (15 g). **c.** SPLAT-DMDS (1 g) + Tomato leaves (15 g) vs Tomato leaves (15 g). An * indicates a significant difference at $P < 0.05$, chi-square test for goodness-of-fit

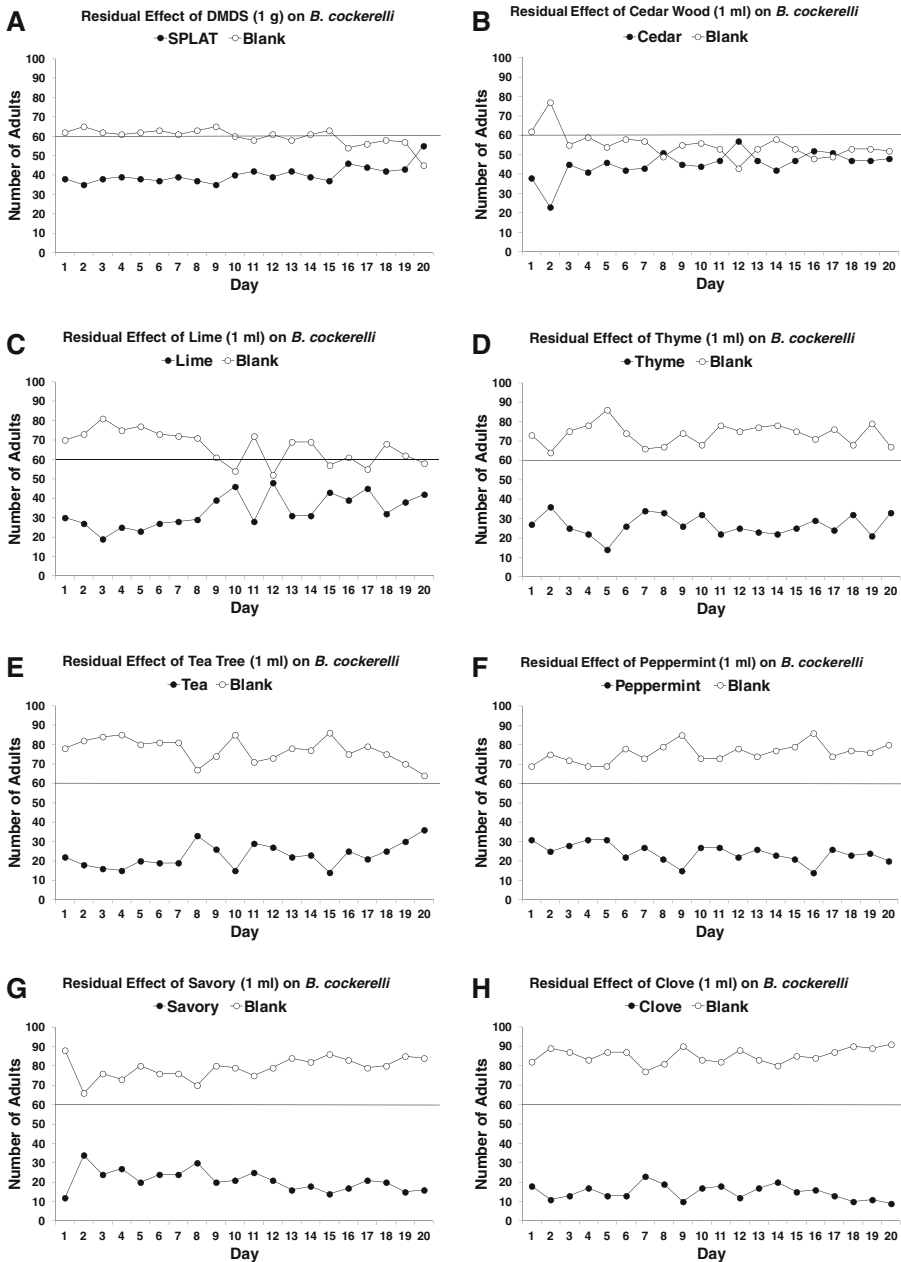


Fig. 3 Residual effect of SPLAT-DMDS (1 g) and seven essential oils (1 ml) on *B. cockerelli* adults for 20 days. Sixty or more adults in the blank arm indicate a significant repellent effect of the compound tested in a given day as denoted by the horizontal line ($P < 0.05$, chi-square test for goodness-of-fit)

average repellency was 54.9 % (Fig. 3b). Lime oil significantly repelled psyllids for the first 9 days, and on days 11, 13–14, 16 and 18–19, with an average repellency of 66.5 % (Fig. 3c). The residual effect of thyme oil was significant throughout the 20-day period. The repellency ranged between 64 and 86 % with an average of 73.5 %

Table 3 Repellent effect of various essential oils on *B. cockerelli* adults

Oil	Dose (μl)	Potato Psyllid Response (%) ^a		χ^2	<i>P</i>
		Blank	Oil		
Patchouli	2,000	58	42	2.56	0.1096
Patchouli	1,000	58	42	2.56	0.1096
Patchouli	500	55	45	1	0.3173
Patchouli	100	58	42	2.56	0.1096
Patchouli	10	54	46	0.64	0.4237
Patchouli	1	52	48	0.16	0.6892
Cassia	2,000	69	31	14.44	0.0001 ^b
Cassia	1,000	54	46	0.64	0.4237
Cassia	500	65	35	9	0.0027 ^b
Cassia	100	57	43	1.96	0.1615
Cassia	10	58	42	2.56	0.1096
Cassia	1	52	48	0.16	0.6892
Garlic	2,000	63	37	6.76	0.0093 ^b
Garlic	1,000	74	26	23.04	0 ^b
Garlic	500	66	34	10.24	0.0014 ^b
Garlic	100	62	38	5.76	0.0164 ^b
Garlic	10	55	45	1	0.3173
Garlic	1	58	42	2.56	0.1096
Celery	2,000	70	30	16	0.0001 ^b
Celery	1,000	68	32	12.96	0.0003 ^b
Celery	500	67	33	11.56	0.0007 ^b
Celery	100	61	39	4.84	0.0278 ^b
Celery	10	47	53	0.36	0.5485
Celery	1	52	48	0.16	0.6892
Rosemary	2,000	70	30	16	0.0001 ^b
Rosemary	1,000	67	33	11.56	0.0007 ^b
Rosemary	500	62	38	5.76	0.0164 ^b
Rosemary	100	69	31	14.44	0.0001 ^b
Rosemary	10	58	42	2.56	0.1096
Rosemary	1	59	41	3.24	0.0719
Clove	2,000	75	25	25	0 ^b
Clove	1,000	72	28	19.36	0 ^b
Clove	500	78	22	31.36	0 ^b
Clove	100	70	30	16	0.0001 ^b
Clove	10	72	28	19.36	0 ^b
Clove	1	51	49	0.04	0.8415
Clove ^c	500	80	20	36	0 ^b
Clove ^d	500	84	16	46.24	0 ^b
Peppermint	2,000	78	22	31.36	0 ^b
Peppermint	1,000	75	25	25	0 ^b

Table 3 (continued)

Oil	Dose (μl)	Potato Psyllid Response (%) ^a		χ^2	<i>P</i>
		Blank	Oil		
Peppermint	500	73	27	21.16	0 ^b
Peppermint	100	75	25	25	0 ^b
Peppermint	10	69	31	14.44	0.0001 ^b
Peppermint	1	53	47	0.36	0.5485
Cedar Wood	2,000	82	18	40.96	0 ^b
Cedar Wood	1,000	78	22	31.36	0 ^b
Cedar Wood	500	78	22	31.36	0 ^b
Cedar Wood	100	79	21	33.64	0 ^b
Cedar Wood	10	67	33	11.56	0.0007 ^b
Cedar Wood	1	62	38	5.76	0.0164 ^b
Thyme	2,000	73	27	21.16	0 ^b
Thyme	1,000	75	25	25	0 ^b
Thyme	500	72	28	19.36	0 ^b
Thyme	100	67	33	11.56	0.0007 ^b
Thyme	10	70	30	16	0.0001 ^b
Thyme	1	68	32	12.96	0.0003 ^b
Tea Tree	2,000	76	24	27.04	0 ^b
Tea Tree	1,000	70	30	16	0.0001 ^b
Tea Tree	500	74	26	23.04	0 ^b
Tea Tree	100	71	29	17.64	0 ^b
Tea Tree	10	70	30	16	0.0001 ^b
Tea Tree	1	63	37	6.76	0.0093 ^b
Lime	2,000	81	19	38.44	0 ^b
Lime	1,000	80	20	36	0 ^b
Lime	500	82	18	40.96	0 ^b
Lime	100	76	24	27.04	0 ^b
Lime	10	78	22	31.36	0 ^b
Lime	1	75	25	25	0 ^b
Savory	2,000	71	29	17.64	0 ^b
Savory	1,000	74	26	23.04	0 ^b
Savory	500	74	26	23.04	0 ^b
Savory	100	72	28	19.36	0 ^b
Savory	10	81	19	38.44	0 ^b
Savory	1	65	35	9	0.0027 ^b

^a 100 psyllid adults tested in each trial^b Significantly more *B. cockerelli* adults preferred the blank arm ($P < 0.05$)^c Only *B. cockerelli* male adults^d Only *B. cockerelli* female adults

(Fig. 3d). Tea tree oil also showed a significant repellency for all 20 days. The percentage of repellency ranged between 64 and 86 % and the average was 77.3 % (Fig. 3e). Peppermint oil significantly repelled *B. cockerelli* adults for all 20 days. The percentage of repellency ranged between 69 and 86 % and the average was 75.8 % (Fig. 3f). Similarly, the residual effect of savory oil on psyllids was significant for 20 days. The percentage of repellency ranged between 66 and 88 % and the average was 79.1 % (Fig. 3g). Clove oil also significantly repelled *B. cockerelli* adults for the 20-day period. The percentage of repellency ranged between 77 and 91 % and the average was 85.3 %. Clove was the only oil that repelled above 90 % of the adults (Fig. 3h).

Essential Oils with Leaves In these experiments, the response of *B. cockerelli* adults to various oils in conjunction with tomato leaves (10 g) was tested against blank arms. Cedar wood oil in the presence of tomato leaves did not repel adults significantly at any dose (1, 10, 100 and 500 μ l) (Table 4). The oils clove and tea tree significantly repelled psyllids at two doses (100 and 500 μ l). Peppermint, thyme, lime and savory significantly repelled *B. cockerelli* adults at three of the four doses (10, 100 and 500 μ l) (Table 4).

In addition, the response of *B. cockerelli* adults to different oils in combination with tomato leaves (10 g) versus just tomato leaves (10 g) was measured. Cedar wood oil did not repel adults significantly at any dose (1, 10, 100 and 500 μ l). Peppermint, thyme, clove, tea tree, lime and savory significantly repelled *B. cockerelli* adults at all doses except at 1 μ l (Table 5).

Discussion

The first objective of this study was to find out if the olfactometer system was an effective tool in eliciting and quantifying a behavioral response of *B. cockerelli* adults to odors. Releasing 50 adults proved to be effective. In addition, their preferences were independent of males or females tested separately or together. In similar experiments with a Y-tube olfactometer using guava leaf volatiles and the Asian citrus psyllid, *D. citri*, the preference of males and females were similar (Zaka et al. 2010). Thus, the data available suggest that gender separation is not necessary for olfactometer trials with at least some species of psyllids. We note that detached leaves with wilting/drought induced emission could be more attractive than intact plants and therefore partly override the repellent effect of tested plant essential oil. Thus, our data may be somewhat conservative.

We believe that DMDS has sufficient potential to initiate field tests for the potato psyllid. DMDS had a significant repellent effect on *B. cockerelli* adults at all doses tested ranging from 62 to 81 %. A repellent effect of DMDS on adults was also observed when it was tested (either in isolation or in combination with plant leaves) versus potato or tomato leaves. This demonstrated that DMDS significantly repelled *B. cockerelli* adults and continued to repel them even in the presence of potato or tomato leaves. If DMDS is effective in the field, the existing registrations on other food crops should expedite permitting on potatoes, peppers, and tomatoes.

Several of the essential oils also have potential for practical application. Of the twelve essential plant oils tested against ‘blanks’, cedar wood, lime, savory,

Table 4 Repellent effect on *B. cockerelli* adults of essential oils along with tomato leaves (10 g) against 'blank' arms in a Y-tube olfactometer system

Oil	Dose (μl)	Potato Psyllid Response (%) ^a		χ^2	P
		Blank	Oil + L		
Cedar Wood	500	54	46	0.64	0.4237
Cedar Wood	100	53	47	0.36	0.5485
Cedar Wood	10	54	46	0.64	0.4237
Cedar Wood	1	55	45	1	0.3173
Clove	500	86	14	51.84	0 ^a
Clove	100	63	37	6.76	0.0093 ^a
Clove	10	45	55	1	0.3173
Clove	1	58	42	2.56	0.1096
Tea Tree	500	82	18	40.96	0 ^a
Tea Tree	100	81	19	38.44	0 ^a
Tea Tree	10	51	49	0.04	0.8415
Tea Tree	1	49	51	0.04	0.8415
Peppermint	500	74	26	23.04	0 ^a
Peppermint	100	72	28	19.36	0 ^a
Peppermint	10	61	39	4.84	0.0278 ^a
Peppermint	1	45	55	1	0.3173
Thyme	500	85	15	49	0 ^a
Thyme	100	74	26	23.04	0 ^a
Thyme	10	75	25	25	0 ^a
Thyme	1	56	44	1.44	0.2301
Lime	500	74	26	23.04	0 ^a
Lime	100	67	33	11.56	0.0007 ^a
Lime	10	65	35	9	0.0027 ^a
Lime	1	59	41	3.24	0.0719
Savory	500	91	9	67.24	0 ^a
Savory	100	84	16	46.24	0 ^a
Savory	10	62	38	5.76	0.0164 ^a
Savory	1	58	42	2.56	0.1096

L = tomato leaves (10 g)

^a Significantly more *B. cockerelli* adults preferred the blank arm ($P < 0.05$)

thyme and tea tree had a repellent effect on *B. cockerelli* adults at all doses. The oils of clove and peppermint also had a repellent effect on adults at all doses except at the lowest dose (1 μl). However, when leaves were present in both Erlenmeyer flasks and the oil was the differential factor, cedar wood oil did not repel psyllids at any of the doses tested. Thus, this material would have much less potential for use in a field or greenhouse. The other effective essential oils repelled the psyllids for a long enough period (at least 20 d) to justify further tests.

Table 5 Repellent effect on *B. cockerelli* adults of essential oils along with tomato leaves (10 g) against tomato leaves (10 g) in a Y-tube olfactometer system

Oil	Dose (μl)	Potato Psyllid Response (%) ^a		χ^2	P
		L	Oil + L		
Cedar Wood	500	48	52	0.16	0.6892
Cedar Wood	100	47	53	0.36	0.5485
Cedar Wood	10	45	55	1	0.3173
Cedar Wood	1	46	54	0.64	0.4237
Clove	500	87	13	54.76	0 ^a
Clove	100	87	13	54.76	0 ^a
Clove	10	62	38	5.76	0.0164 ^a
Clove	1	53	47	0.36	0.5485
Tea Tree	500	88	12	57.76	0 ^a
Tea Tree	100	73	27	21.16	0 ^a
Tea Tree	10	62	38	5.76	0.0164 ^a
Tea Tree	1	55	45	1	0.3173
Peppermint	500	70	30	16	0.0001 ^a
Peppermint	100	74	26	23.04	0 ^a
Peppermint	10	63	37	6.76	0.0093 ^a
Peppermint	1	53	47	0.36	0.5485
Thyme	500	80	20	36	0 ^a
Thyme	100	84	16	46.24	0 ^a
Thyme	10	77	23	29.16	0 ^a
Thyme	1	45	55	1	0.3173
Lime	500	74	26	23.04	0 ^a
Lime	100	67	33	11.56	0.0007 ^a
Lime	10	67	33	11.56	0.0007 ^a
Lime	1	46	54	0.64	0.4237
Savory	500	87	13	54.76	0 ^a
Savory	100	74	26	23.04	0 ^a
Savory	10	62	38	5.76	0.0164 ^a
Savory	1	56	44	1.44	0.2301

L = tomato leaves (10 g)

^a Significantly more *B. cockerelli* adults preferred the arm with leaves (L) ($P < 0.05$)

In order to be useful in the field or greenhouse, these materials must work even in the presence of plants. If DMDS and the oils of thyme, tea tree, peppermint, savory and clove prove as effective in the field as in these laboratory experiments, they could be useful in an IPM program for *B. cockerelli*. However, even though some compounds appear to have potential and can be registered in some countries for organic production, possible effects on beneficial insects, longevity of materials in the field, delivery systems, and cost/benefit will need to be determined before any deployment in agricultural systems.

Acknowledgments We thank Greg Kund, Beatriz Gonzalez, Bill Carson, Steve McElfresh, Deborah De La Riva, Kristen Hladun, Christina Mogren, Nichole Drew, Sammuell Gilbert, Khoa Tran (Chris), Kristine Gilbert and Lindsay Johnson for helping in different aspects of this study. Agenor Mafra-Neto and Lyndsie Stoltman (ISCA Technologies, Riverside, CA). This research was partially funded by a USDA SCRI grant (2009-34381-20036), a USDA RAMP grant (2009-51101-05892) and the California Potato Board. Mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the U.S. Department of Agriculture or the University of California-Riverside.

References

- Adorjan B, Buchbauer G (2010) Biological properties of essential oils: an updated review. *Flavour Frag J* 25:407–426
- Al-Jabr AM, Whitney SC (2007) Trapping tomato psyllid, *Bactericera cockerelli* (Sulc) (Hemiptera: Psyllidae), in greenhouses. *Southwest Entomol* 32:25–30
- Borges M, Mori K, Costa MLM, Sujii ER (1998) Behavioural evidence of methyl-2,6,10-trimethyltridecanoate as a sex pheromone of *Euschistus heros* (Het., Pentatomidae). *J Appl Entomol* 122:335–338
- Davidson MM, Perry NB, Larsen L, Green VC, Butler RC, Teulon DAJ (2008) 4-Pyridyl Carbonyl compounds as thrips lures: effectiveness for Western flower thrips in Y-tube bioassays. *J Agric Food Chem* 56:6554–6561
- Egigu MC, Ibrahim MA, Yahya A, Holopainen JK (2010) Yeheb (*Cordeauxia edulis*) extract deters feeding and oviposition of *Plutella xylostella* and attracts its natural enemy. *Biocontrol* 55:613–624
- Gharalari AH, Nansen C, Lawson DS, Gilley J, Munyaneza JE, Vaughn K (2009) Knockdown, mortality, repellency, and residual effects of insecticides for control of *Bactericera cockerelli* adult (Hemiptera: Psyllidae). *J Econ Entomol* 102:1032–1038
- Gökçe A, Steilinski LL, Whalon ME (2005) Behavioral and Electrophysiological Responses of Leafroller Moths to Selected Plant Extracts. *Environ Entomol* 34:1426–1432
- Goolsby JA, Adamczyk J, Bextine B, Lin D, Munyaneza JE, Bester G (2007) Development of an IPM program for management of the potato psyllid to reduce incidence of zebra chip disorder in potatoes. *Subtrop Pl Sci* 59:85–94
- Guedot C, Millar JC, Horton DR, Landholt PJ (2009) Identification of a sex attractant pheromone for male winterform pear psylla, *Cacopsylla pyricola*. *J Chem Ecol* 35:1437–1447
- Hansen AK, Trumble JT, Stouthamer R, Paine TD (2008) A new Huanglongbing species, “*Candidatus Liberibacter psyllaurosus*”, found to infect tomato and potato, is vectored by the psyllid *Bactericera cockerelli* (Sulc). *Appl Environ Microbiol* 74:5862–5865
- Hardie J, Holyoak M, Nicholas J, Nottingham SF, Pickett JA, Wadhams LJ, Woodcock CM (1990) Aphid sex pheromone components: age-dependent release by females and species-specific male response. *Chemoecology* 1:63–68
- Isman MB (2006) Botanical insecticides, deterrents, and repellents in modern agriculture and an increasingly regulated world. *Ann Rev Entomol* 51:45–66
- Kainulainen P, Nissinen A, Piirainen A, Tiilikkala K, Holopainen JK (2002) Essential oil composition in leaves of carrot varieties and preference of specialist and generalist sucking insect herbivores. *Agr Forest Entomol* 4:211–216
- Koschier EH, Sedy KA (2003) Labiate essential oils affecting host selection and acceptance of *Thrips tabaci* Lindeman. *Crop Prot* 22:929–934
- Koschier EH, de Kogel WJ, Visser JH (2000) Assessing the attractiveness of volatile plant compounds to western flower thrips *Frankliniella occidentalis*. *J Chem Ecol* 26:2643–2655
- Liefting LW, Perez-Egusquiza XC, Clover GRG (2008) A new ‘*Candidatus Liberibacter*’ species in *Solanum tuberosum* in New Zealand. *Plant Dis* 92:1474
- Liefting LW, Southerland PW, Ward LI, Paice KL, Weir BS, Clover GRG (2009) A new “*Candidatus Liberibacter*” species associated with diseases of solanaceous crops. *Plant Dis* 93:208–214
- Liu DG, Trumble JT (2005) Interactions of plant resistance and insecticides on the development and survival of *Bactericera cockerelli* [Sulc] (Homoptera: Psyllidae). *Crop Prot* 24:111–117
- Liu DG, Trumble JT (2006) Ovipositional preferences, damage thresholds, and detection of the tomato-potato psyllid *Bactericera cockerelli* (Homoptera: Psyllidae) on selected tomato accessions. *Bull Ent Res* 96:197–204

- Liu DG, Trumble JT (2007) Comparative fitness of invasive and native populations of the potato psyllid *Bactericera cockerelli*. *Entomol Exp Appl* 123:35–42
- Mann RS, Rouseff RL, Smoot JM, Castle WS, Stelinski LL (2011) Sulfur volatiles from *Allium* spp. affect Asian citrus psyllid, *Diaphorina citri* Kuwayama (Hemiptera: Psyllidae), response to citrus volatiles. *Bull Ent Res* 101:89–97
- Moharramipour S, Taghizadeh A, Meshkatsadat MH, Talebi AA, Fathipour Y (2008) Repellent and fumigant toxicity of essential oil from *Thymus persicus* against *Tribolium castaneum* and *Callosobruchus maculatus*. *Commun Agric Appl Biol Sci* 73:639–642
- Munyanza JE (2010) Psyllids as vectors of emerging bacterial diseases of annual crops. *Southwest Entomol* 35:471–477
- Munyanza JE, Crosslin JM, Upton EJ (2007a) Association of *Bactericera cockerelli* (Homoptera: Psyllidae) with “Zebra Chip”, a new potato disease in Southwestern United States and Mexico. *J Econ Entomol* 100:656–663
- Munyanza JE, Goolsby JA, Crosslin JM, Upton EJ (2007b) Further evidence that Zebra Chip potato disease in the Lower Rio Grande Valley of Texas is associated with *Bactericera cockerelli*. *Subtrop PI Sci* 59:30–37
- Munyanza JE, Buchman JL, Upton JE, Goolsby JA, Crosslin JM, Bester G, Miles GP, Sengoda VG (2008) Impact of different potato psyllid populations on Zebra Chip disease incidence, severity, and potato yield. *Subtrop PI Sci* 60:27–37
- Munyanza JE, Sengoda VG, Crosslin JM, Rosa-Lozano GD, Sanchez A (2009) First report of *Candidatus Liberibacter psyllaurosus* in potato tubers with Zebra Chip disease in Mexico. *Plant Dis* 93:552
- Nehlin G, Valterová I, Borg-Karlson AK (1994) Use of conifer volatiles to reduce injury caused by carrot psyllid, *Triozia apicalis*, Förster (Homoptera, Psylloidea). *J Chem Ecol* 20:771–783
- Nerio LS, Olivero-Verbel J, Stashenko E (2010) Repellent activity of essential oils: a review. *Bioresour Technol* 101:372–378
- Nissinen A, Ibrahim M, Kainulainen P, Tiilikkala K, Holopainen JK (2005) Influence of carrot psyllid (*Triozia apicalis*) feeding or exogenous limonene or methyl jasmonate treatment on composition of carrot (*Daucus carota*) leaf essential oil and headspace volatiles. *J Agric Food Chem* 53:8631–8638
- Onagbola EO, Rouseff RL, Smoot JM, Stelinski LL (2011) Guava leaf volatiles and dimethyl disulphide inhibit response of *Diaphorina citri* Kuwayama to host plant volatiles. *J Appl Entomol* 135:404–414
- Rodriguez-Saona C, Poland TM, Miller JR, Stelinski LL, Grant GG, de Groot P, Buchan L, MacDonald L (2006) Behavioral and electrophysiological responses of the emerald ash borer, *Agrilus planipennis*, to induced volatiles of Manchurian ash, *Fraxinus mandshurica*. *Chemoecology* 16:75–86
- SAS Institute (2003) SAS/STAT user’s guide, version 9.1.3. SAS institute, Cary
- Teulon DAJ, Workman PJ, Thomas KL, Nielsen MC (2009) *Bactericera cockerelli*: incursion, dispersal and current distribution on vegetable crops in New Zealand. *N Z Plant Protection* 62:136–144
- Turlings TCJ, Davison AC, Tamo C (2004) A six-arm olfactometer permitting simultaneous observation of insect attraction and odour trapping. *Physiol Entomol* 29:45–55
- Valterová I, Nehlin G, Borg-Karlson AK (1997) Host plant chemistry and preferences in egg-laying *Triozia apicalis* (Homoptera, Psylloidea). *Biochem Syst Ecol* 25:477–491
- Yang NW, Li AL, Wan FH, Liu WX, Johnson D (2010a) Effects of plant essential oils on immature and adult sweetpotato whitefly, *Bemisia tabaci* biotype B. *Crop Prot* 29:1200–1207
- Yang XB, Zhang YM, Hua L, Peng LN, Munyanza JE, Trumble JT, Liu TX (2010b) Repellency of selected biorational insecticides to potato psyllid, *Bactericera cockerelli* (Hemiptera: Psyllidae). *Crop Prot* 29:1320–1324
- Zaka SM, Zeng XN, Holford P, Beattie GAC (2010) Repellent effect of guava leaf volatiles on settlement of adults of citrus psylla, *Diaphorina citri* Kuwayama, on citrus. *Insect Sci* 17:39–45